SIMULTANEOUS DETECTION OF STRUCTURAL AND NUMERICAL CHROMOSOME ABERRATIONS IN HUMAN SPERM BY MULTI-COLOR FISH.

Paul Van Hummelen*, Xiu Lowe, Andrew J Wyrobek

Biology and Biotechnology Research Program, Lawrence Livermore National Lab., PB 808, Livermore, CA, 94550

Structural and numerical chromosome aberrations transmitted via germ cells are major contributors to pregnancy loss and congenital abnormalities. Although the parental origin of autosomal aneuploidies are predominantly maternal, the contribution of the father is substantial in sex chromosome aneuploidies and de novo structural aberrations. Therefore there is a general concern about the hazards of paternal exposure to mutagenic agents via environment, occupation or lifestyle. Recently, methods have been developed to assess chromosomal damage in sperm using fluorescence in situ hybridization (FISH). The sperm nucleus presents a special challenge for in situ hybridization because of the tight packaging of DNA which interferes with penetration by DNA probes.

To detect aneuploidy, methodologies were developed that progressively increased the number of simultaneous detection of centromeric specific DNA probes in sperm. In the most recent method, up to four probes could simultaneously be labeled in different colors. The chromosomes under investigation in our laboratory are X, Y, 16, 18, and 21 representing the majority of aneuploid abortions or offspring in humans. Since most of the mutagenic agents are chromosome breaking agents an additional method was developed to assess structural aberrations in chromosome 1p, using probes labeling both the centromere and its telomere in different colors.

Baseline frequencies for healthy donors have been determined and excellent agreements were obtained with the human-sperm/hamster-egg technique for sperm cytogenetics indicating the validity of the hybridization measurements for both structural aberrations and aneuploidy detection. These assays were used to detect increases of structural and numerical aberrations in Hodgkin's disease patients undergoing chemotherapy and in cigarette smokers. We propose to apply these newly developed methods to detect and characterize effects of exposure to mutagens and to evaluate host factors that may predispose individuals to produce chromosomally defective sperm.

[Work was performed under the auspices of the U.S. DOE by the Lawrence Livermore National Laboratory under contract W-7405-ENG-48]